

Potentially Actionable Suspect Sample (PASS) System – *Phytophthora ramorum*
April 2006
(Host List Updated September 11, 2006)

The purpose of this document is to create a submission system for APHIS provisionally approved laboratories or laboratories with identification authority. This system will reduce the time needed to determine if *P. ramorum* is present in samples by limiting the required number of samples to be submitted to APHIS for confirmation and insure clarity on what samples need to be submitted.

Introduction:

The current *P. ramorum* diagnostic policy is described in the Federal Emergency Order effective January 10, 2005, (see, www.aphis.usda.gov/ppq/ispm/pramorum/). The establishment of provisionally approved laboratories for PCR diagnostics and the issuing of identification authority for cultures are intended to significantly reduce the time needed to determine if *P. ramorum* is present in samples. The proposed Potentially Actionable Suspect Sample System (PASS) is two fold. While ensuring Federal determination of key samples, this process will also eliminate the need to submit redundant presumptive positive cultures or DNA from samples, reducing the number of samples requiring confirmation by APHIS/PPQ laboratories and thereby reducing the time interval between sampling and final identification.

Purpose:

The purpose of this document is to provide guidance to APHIS provisionally approved laboratories or diagnosticians with identification authority regarding which *P. ramorum* presumptive positives must be submitted to APHIS for confirmation.

Definitions:

Associated plants:

Associated plants are those reported found naturally infected and from which *P. ramorum* has been cultured and/or detected using PCR (Polymerase Chain Reaction). For each of these, traditional Koch's postulates have not yet been completed or documented and reviewed. See Appendix 1.

Confirmed Positive:

The test result on a presumptive positive that *P. ramorum* is present based on DNA testing or culture morphology. This confirmation would be conducted by APHIS in the case of PASS samples or by the provisionally approved lab or diagnosticians with

identification authority in the case of non-PASS samples.

HAP:

Host and associated host plants listed on the official APHIS List of Regulated Hosts and Plants Associated with *Phytophthora ramorum*.

Host plants:

Naturally infected plants verified with completion, documentation, review and acceptance of traditional Koch's postulates and listed in the "APHIS List of Regulated Hosts and Plants Associated with *Phytophthora ramorum*".

Identification Authority:

Authority to confirm the presence of *P. ramorum* issued by the APHIS National Identification Services – National Mycologist, Beltsville to diagnosticians that have demonstrated proficiency in identifying *P. ramorum* in culture.

Provisional Approval:

Authority to perform approved PCR diagnostics for *P. ramorum* issued by the APHIS National Plant Germplasm and Biotechnology Laboratory, Beltsville to laboratories that have demonstrated proficiency in detecting and identifying *P. ramorum* DNA.

Potentially Actionable Suspect Sample (PASS):

A presumptive positive *P. ramorum* sample diagnosed or identified by a provisionally approved laboratory or diagnostician with identification authority that would require confirmatory testing by an official APHIS Laboratory due to the nature of the plant sampled and the necessity for Federal confirmation.

Presumptive Positive:

Such a result may require confirmatory testing if the sample is a PASS sample.

Trace Forward (TF) Plants:

Plants identified on a trace forward list as being potentially infected with *P. ramorum*.

Trace Forward (TF) Site:

Any location that received potentially infected plants from a confirmed infested source nursery; including residential or commercial landscapes.

Trace Back (TB) Plants:

Plants identified on a trace back list as being potentially infected with *P. ramorum*.

Trace Back (TB) Site:

Any source location, including residential or commercial landscapes, which shipped presumptive, confirmed or potentially infected plants.

Routing for Samples:

If the sample is an Initial Presumptive Positive from a:	Then the sample is a:
National survey site	PASS Sample and must be sent to an APHIS Laboratory for confirmation ASAP, within 2 weeks.
Compliance Inspection site	
Cleanliness Inspection site	
Certification Inspection site	
TF plant at a TF site that ships interstate	
TF plant at a TF site in a state other than the source site	
(TB) site	
Any unusual or unexpected detection or one not otherwise covered above*	
If the sample is a subsequent Presumptive Positive from:	Then the sample is:
National survey site	Not a PASS sample if the sample is covered by previous confirmation of the PASS sample.
Compliance Inspection site	
Cleanliness Inspection site	
Certification Inspection site	
TF plant at a TF site that does not ship interstate and is in the same state as the source site	
TB site (originating source)	
If the sample is an Initial Presumptive Positive or a Subsequent Presumptive Positive from:	Then the sample is a:
Any TF site where the sample is from any plant not part of the TF shipments(s)	PASS Sample and must be sent to an APHIS Laboratory for confirmation ASAP, within two weeks.
Any sample that will require Federal regulatory action	
Any previously undescribed or unknown host(s)*	
Any new host or host not previously reported found in the US*	
Any environmental location outside the current quarantined counties, including home owner's yards, natural landscape or forest location(s) whether or not associated with a positive nursery*	

*Do to the potential impact and importance of these samples, these samples will also require that a second official sample be collected, processed, and Federally confirmed. Inconsistent results from the two samples may require additional sampling and testing to be determined on a case-by-case basis.

The PASS sample may be a single sample or may be several samples collected at the same time. In any instance where a culture of *P. ramorum* has been obtained, confirmatory testing of DNA extracted from regulatory samples is not required, although confirmation of the ID of the culture is required. It should also be understood that following the mitigation of the disease as specified in the APHIS protocols, and the nursery is again seeking certification, or the site has been declared free of *P. ramorum* or official control, the process is re-started.

Samples from laboratories that are not provisionally approved or laboratories not granted with identification authority must be treated in the manner described in the emergency Federal order effective January 10, 2005. That is, all DNA extractions of ELISA positive plants and cultures must be submitted to APHIS for determination.

This process applies to each nursery certification, e.g., if a nursery was determined to be positive, mitigation measures were undertaken as specified in the APHIS protocols, and the nursery is again seeking certification, the PASS process begins again. The above decision table is for provisionally approved laboratories or diagnosticians with identification authority. Samples from laboratories that are not provisionally approved or to which identification authority has not been granted must be treated in the manner described in the Federal Emergency Order (effective January 10, 2005) and all DNA extractions and cultures must be submitted to APHIS for final determination.

Appendix 1

APHIS List of Regulated Hosts and Plants Associated with *Phytophthora ramorum*

(Revision dated 11 September 2006)

This list is continually being updated.

The most current version is posted at: <http://www.aphis.usda.gov/ppq/ispm/pramorum>

Proven Hosts Regulated for *Phytophthora ramorum*

Scientific Name (47)	Common Name(s)	Notes
<i>Acer macrophyllum</i>	Bigleaf maple	
<i>Acer pseudoplatanus</i>	Planetree maple	Koch's postulates completed
<i>Aesculus hippocastanum</i>	Horse chestnut	Koch's postulates completed
<i>Adiantum aleuticum</i>	Western maidenhair fern	
<i>Adiantum jordanii</i>	California maidenhair fern	
<i>Aesculus californica</i>	California buckeye	
<i>Arbutus menziesii</i>	Madrone	
<i>Arctostaphylos manzanita</i>	Manzanita	
<i>Calluna vulgaris</i>	Scotch heather	
<i>Camellia</i> spp.	Camellia - all species, hybrids and cultivars	
<i>Castanea sativa</i>	Sweet chestnut	
<i>Fagus sylvatica</i>	European beech	
<i>Frangula californica</i> (= <i>Rhamnus californica</i>)	California coffeeberry	
<i>Frangula purshiana</i> (= <i>Rhamnus purshiana</i>)	Cascara	
<i>Fraxinus excelsior</i>	European ash	
<i>Griselinia littoralis</i>	Griselinia	
<i>Hamamelis virginiana</i>	Witch hazel	
<i>Heteromeles arbutifolia</i>	Toyon	
<i>Kalmia latifolia</i>	Mountain laurel	
<i>Lithocarpus densiflorus</i>	Tanoak	
<i>Lonicera hispidula</i>	California honeysuckle	
<i>Laurus nobilis</i>	Bay laurel	Koch's postulates completed
<i>Maianthemum racemosum</i> (= <i>Smilacina racemosa</i>)	False Solomon's seal	

<i>Michelia doltsopa</i>	Michelia	Koch's postulates completed
<i>Parrotia persica</i>	Persian ironwood	
<i>Photinia fraseri</i>	Red tip photinia	
<i>Pieris floribunda</i> and <i>Pieris floribunda x japonica</i> & all hybrids of <i>P. floribunda</i>	Mountain Andromeda	
<i>Pieris formosa</i> and <i>P. formosa x japonica</i> & all hybrids of <i>P. formosa</i>	Himalaya Andromeda	
<i>Pieris japonica</i> & all hybrids of <i>P. japonica</i>	Japanese Pieris	
<i>Pseudotsuga menziesii</i> var. <i>menziesii</i> & all nursery grown <i>P. menziesii</i>	Douglas fir	
<i>Quercus agrifolia</i>	Coast live oak	
<i>Quercus chrysolepis</i>	Canyon live oak	
<i>Quercus cerris</i>	European turkey oak	
<i>Quercus falcata</i>	Southern red oak	
<i>Quercus ilex</i>	Holm oak	
<i>Quercus kelloggii</i>	California black oak	
<i>Quercus parvula</i> var. <i>shrevei</i> & all nursery grown <i>Q. parvula</i>	Shreve's oak	
<i>Rhododendron spp.</i>	Rhododendron (including azalea) – all species, hybrids and cultivars	
<i>Rosa gymnocarpa</i>	Wood rose	
<i>Salix caprea</i>	Goat willow	
<i>Sequoia sempervirens</i>	Coast redwood	
<i>Syringa vulgaris</i>	Lilac	
<i>Taxus baccata</i>	European yew	
<i>Trientalis latifolia</i>	Western starflower	
<i>Umbellularia californica</i>	California bay laurel, pepperwood, Oregon myrtle	
<i>Vaccinium ovatum</i>	Evergreen huckleberry	
<i>Viburnum spp.</i>	Viburnum – all species, hybrids and cultivars	

Plants Associated with *Phytophthora ramorum*

(These are regulated only as nursery stock)

Scientific Name (58)	Common Name, Date & Source of Report	Notes
<i>Abies concolor</i>	White fir – Oct 05 (1)	
<i>Abies grandis</i>	Grand fir – June 03 (1)	
<i>Abies magnifica</i>	Red fir – Jan 06 (7)	
<i>Acer circinatum</i>	Vine maple – Feb 06 (5)	
<i>Acer davidii</i>	Striped bark maple – Jan 06 (9)	
<i>Acer laevigatum</i>	Evergreen Maple – Aug 05 (3)	
<i>Arbutus unedo</i>	Strawberry tree – Dec 02 (7)	
<i>Arctostaphylos columbiana</i>	Manzanita – Feb 06 (5)	
<i>Ardisia japonica</i>	Ardisia – Jan 06 (9)	
<i>Calycanthus occidentalis</i>	Spicebush – May 05 (5)	
<i>Castanopsis orthacantha</i>	Castanopsis - Aug 06 (3)	New listing - Reported found in the UK
<i>Ceanothus thyrsiflorus</i>	Blueblossom – April 06 (5)	
<i>Cinnamomum camphora</i>	Camphor tree – May 06 (3)	
<i>Clintonia andrewsiana</i>	Andrew's clintonia bead lily – May 04 (5)	
<i>Cornus kousa x Cornus capitata</i>	Cornus Norman Haddon – Aug 06 (3)	New listing - Reported found in the UK
<i>Corylus cornuta</i>	California hazelnut – Dec 02 (5)	
<i>Distylium myricoides</i>	Myrtle-leaved Distylium – Jul 06 (9)	New listing - Reported found in Canada
<i>Drimys winteri</i>	Winter's bark – July 04 (3)	
<i>Dryopteris arguta</i>	California wood fern – May 04 (5)	
<i>Eucalyptus haemastoma</i>	Scribbly gum – Aug 06 (3)	New listing - Reported found in the UK
<i>Euonymus kiautschovicus</i>	Spreading euonymus – Jan 06 (9)	
<i>Fraxinus latifolia</i>	Oregon ash – Aug 05 (5)	
<i>Gaultheria shallon</i>	Salal, Oregon wintergreen – Jan 06 (9)	
<i>Hamamelis x intermedia</i> (<i>H. mollis</i> & <i>H. japonica</i>)	Hybrid witchhazel – Jan 06 (9)	

<i>Hamamelis mollis</i>	Chinese witchhazel – Jan 05 (3)	
<i>Ilex purpurea</i>	Oriental holly – Jul 06 (9)	New listing - Reported found in Canada
<i>Kalmia angustifolia</i>	Sheep laurel – May 06 (3)	
<i>Leucothoe axillaris</i>	Fetterbush, dog hobble – Jan 06 (9)	
<i>Leucothoe fontanesiana</i>	Drooping leucothoe - Oct 03 (3)	
<i>Loropetalum chinense</i>	Loropetalum – Jul 06 (9)	New listing - Reported found in Canada
<i>Manglietia insignis</i>	Red lotus tree – Aug 06 (9)	New listing - Reported found in Canada
<i>Magnolia grandiflora</i>	Southern magnolia – Jan 06 (9)	
<i>Magnolia stellata</i>	Star magnolia – Jan 05 (3)	
<i>Magnolia x loebneri</i>	Loebner magnolia – Jan 05 (3)	
<i>Magnolia x soulangeana</i>	Saucer magnolia – Jan 05 (3)	
<i>Michelia maudiae</i>	Michelia – Jan 06 (9)	
<i>Michelia wilsonii</i>	Michelia – Jan 06 (9)	
<i>Nerium oleander</i>	Oleander – June 06 (1)	
<i>Nothofagus obliqua</i>	Roble beech – Dec 04 (3)	
<i>Osmorhiza berteroi</i>	Sweet Cicely – Aug 05 (5)	
<i>Osmanthus decorus</i> (≡ <i>Phillyrea decora</i> ; ≡ <i>P. vilmoriniana</i>)	Osmanthus – Jan 06 (9)	
<i>Osmanthus fragrans</i>	Sweet olive – June 06 (1)	
<i>Osmanthus heterophyllus</i>	Holly olive – June 06 (1)	
<i>Parakmeria lotungensis</i>	Eastern joy lotus tree – Jul 06 (9)	New listing - Reported found in Canada
<i>Pittosporum undulatum</i>	Victorian box – Dec 02 (6)	
<i>Prunus lusitanica</i>	Portuguese laurel cherry – Jan 06 (9)	
<i>Pyracantha koidzumii</i>	Formosa firethorn – Apr 04 (9)	
<i>Quercus acuta</i>	Japanese evergreen oak – May 06 (3)	
<i>Quercus petraea</i>	Sessile oak – Aug 05 (3)	
<i>Quercus rubra</i>	Northern red oak – Nov 03 (8)	
<i>Rosa</i> (specific cultivars) Royal Bonica (tagged:	Hybrid roses – Jan 06 (9)	Revised listing - Note that these are specific registered cultivars which can be identified by the

“MEImodac”)		listed tags
Pink Meidiland (tagged: “MEIpoque”)		
Pink Sevillana (tagged: “MEIgeroka”)		
<i>Rosa rugosa</i>	Rugosa rose – Jan 06 (9)	
<i>Rubus spectabilis</i>	Salmonberry – Dec 02 (4)	
<i>Taxus brevifolia</i>	Pacific yew – May 03 (5)	
<i>Taxus x media</i>	Yew – June 05 (8)	
<i>Torreya californica</i>	California nutmeg – Aug 05 (5)	
<i>Toxicodendron diversilobum</i>	Poison oak – Dec 02 (4)	
<i>Vancouveria planipetala</i>	Redwood ivy – Aug05 (5)	

- ¹ California Department of Food and Agriculture, Sacramento, CA
- ² Oregon Department of Agriculture. Salem, OR
- ³ Department for Environment, Food and Rural Affairs, UK
- ⁴ Everett Hanson, Oregon State University, Corvallis, OR
- ⁵ David Rizzo, University of California, Davis, CA
- ⁶ Matteo Garbelotto, University of California, Berkeley, CA
- ⁷ Gary Chastagner, Washington State University, Puyallup, WA
- ⁸ Plant Protection Service, Wageningen, Netherlands
- ⁹ Canadian Food Inspection Agency, Ottawa, Ontario, Canada
- ¹⁰ (Reserved)
- ¹¹ (Reserved)

Rationale for Lists:

Host Plants Regulated for *Phytophthora ramorum*:

Naturally infected associated plants are deemed host plants regulated for *P. ramorum* upon completion, documentation, review and acceptance of traditional Koch's postulates. Details on regulated plants and articles can be found via links to "Phytophthora ramorum 7 CFR 301.92" and "Recent Modifications to Phytophthora ramorum Regulations" at: <http://www.aphis.usda.gov/ppq/ispm/pramorum>

The plants listed in the original Interim Rule dated 14 February 2002 were adapted from a review and evaluation of lists of regulated plants from other regulatory agencies.

Plants Associated with *Phytophthora ramorum*:

Plants associated with *P. ramorum* are naturally infected plants and from which *P. ramorum* has been cultured and/or detected using PCR (Polymerase Chain Reaction). Traditional Koch's postulates have not yet been completed nor documented and reviewed for each of these associated plants. These reports must be documented and reviewed by PPQ before they will be listed.

Regulation at the genus level:

Plants included in either of the above lists may be regulated at the genus level. This will ensure appropriate and effective inspection in quarantine areas, regulated nurseries, and regulated articles to mitigate the spread of *P. ramorum*. An example is when the number of individual species, hybrids, or cultivars listed or to be listed is determined to hinder appropriate and effective inspection or regulation.

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